AGRONOMY (AGRONOMIA)



The residues of *Canavalia ensiformis* and *Crotalaria juncea* after phytoremediation in soil contaminated with sulfentrazone do not result in soil recontamination

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ABSTRACT: Phytoremediation is an alternative for the decontamination of areas that have received intense applications of herbicides. This study aimed to verify the reuse possibility of *Canavalia ensiformis* and *Crotalaria juncea* as green manure and the influence of the vegetation cover after phytoremediation of soil contaminated with sulfentrazone on the growth and development of *Pennisetum glaucum*. Two experiments were carried out in a greenhouse. One of the experiments was conducted with *C. juncea* and the other with *C. ensiformis*, two phytoremediation species of the sulfentrazone herbicide. The experiments were set up in 4 x 3 factorial, with the first factor being four population densities of the phytoremediate species (*C. juncea* - 0, 60, 120 and 240 plants m⁻²; *C. ensiformis* - 0, 10, 20 and 40 plants m⁻²), and the second composed by three sulfentrazone doses (0, 200 and 400 g ha⁻¹). Seventy-five days after emergence (DAE), the plants were collected by separating the aerial parts and roots to determine the sulfentrazone accumulation in the plants by using high performance liquid chromatography. Plant extract was prepared from the vegetable material and applied in pots containing washed sand, followed by the sowing of the indicative plant (*Pennisetum glaucum*). It was evaluated as to its phytotoxicity plant height, aerial part and roots dry matter. *P. glaucum* cultivated in the substrate that received leaf extract from *C. ensiformis* and *C. juncea* cultivated in soils with presence of sulfentrazone did not display any intoxication symptom to this herbicide. The extracts from *Canavalia* ensiformis and *Crotalaria juncea* were not phytotoxic to the millet, indicating that these species can be used as green fertilization after phytoremediation of the sulfentrazone in the soil.

Key words: environmental contamination; phytoextraction; vegetal cover; vegetal extract

A permanência de resíduos de *Canavalia ensiformis* e *Crotalaria juncea* após fitorremediação em solo contaminado com sulfentrazone não resulta em recontaminação do solo

RESUMO: A fitorremediação é uma alternativa para a descontaminação de áreas que receberam intensas aplicações de herbicidas. Objetivou-se com esse estudo verificar a possibilidade de reutilização de *Canavalia ensiformis* e *Crotalaria juncea* como adubo verde e a influência da cobertura vegetal após fitorremediação de solo contaminado com sulfentrazone sobre o crescimento e desenvolvimento de *Pennisetum glaucum*. Foram realizados dois experimentos em casa-de-vegetação. Um dos experimentos foi conduzido com *C. juncea* e outro com *C. ensiformis*, duas espécies fitorremediadoras do herbicida sulfentrazone. Os experimentos foram montados em fatorial 4 x 3, sendo o primeiro fator quatro densidades populacionais das espécies fitorremediadoras (*C. juncea* - 0, 60, 120 e 240 plantas m⁻²; *C. ensiformis* - 0, 10, 20 e 40 plantas m⁻²) e o segundo três doses de sulfentrazone (0, 200 e 400 g ha⁻¹). Setenta e cinco dias após a emergência (DAE) as plantas foram coletadas separando-se a parte aérea e a raiz para determinação da acumulação de sulfentrazone nas plantas por cromatografia líquida de alta eficiência. Com parte do material vegetal foi preparado um extrato vegetal que foi aplicado em potes contendo areia lavada e em seguida foi semeada a planta indicadora (*Pennisetum glaucum*). Esta foi avaliada quanto a fitotoxicidade, altura de plantas, matéria seca da parte aérea e da raiz. O *P. glaucum* cultivado em substrato que recebeu extrato foliar de *C. ensiformis* e *C. juncea* cultivadas em solos com a presença de sulfentrazone, não apresentou nenhum sintoma de intoxicação a esse herbicida. Os extratos de *Canavalia ensiformis* e *Crotalaria juncea* não foram fitotóxicos para o milheto indicando que essas espécies podem ser utilizadas como adubação verde após fitorremediar o sulfentrazone no solo.

Palavras-chave: contaminação ambiental; fitoextração; cobertura vegetal; extrato vegetal

Introduction

There is great concern nowadays regarding the environmental contamination caused by the indiscriminate use of xenobiotics such as herbicides. When the herbicides reach the soil, the processes of redistribution and degradation, which may be extremely short or persist for months or even years, are initiated (Filizola et al., 2002; Stipičević et al., 2015; Wang et al., 2017; Barik et al., 2017).

Among the herbicides that have a long residual period in the soil is the sulfentrazone, recommended for application in preemergence on the control of weeds in the crops of sugarcane, soybean, citrus, coffee, eucalyptus and non-agricultural areas (Rodrigues & Almeida, 2005). Due to its long persistence in the soil, this herbicide can invalidate the growth of sensitive plants for a long time after its application, depending on the applied dose and the edaphoclimatic conditions (Vivian et al., 2006). In addition to being persistent, sulfentrazone is mobile in the soil and has high leaching potential (Paraíba et al., 2003; Martinez et al., 2008, Passos et al., 2015), being able to reach groundwater.

In order to minimize the environmental impact caused by the use of herbicides, especially those with long activity period in the soil, several techniques have been employed. Phytoremediation, which involves the use of plant species for accelerating the detoxification of soils and waters contaminated with toxic compounds (Cunningham et al., 1996), has been a promising option for the efficient treatment of edaphic environments contaminated with herbicides (Pires et al., 2003).

However, in a phytoremediation process, especially with an agricultural approach, it is important that the remediation plants degrade the herbicide so that it is not necessary to remove these plants from the site at the end of the process. In case this proves necessary, the costs with the system can be high, resulting in the lack of interest from the farmers in adopting the technique.

Previous studies proved the efficiency of *Canavalia ensiformis* and *Crotalaria juncea* in remediating sulfentrazone (Madalão et al., 2012a; b; 2013; 2017; Ferraço et al., 2017). In the phytoremediation of soils with this herbicide, the *C. ensiformis* and *C. juncea* species are expected to be reused in green fertilization, making possible the insertion of these two in a program of rotation or succession of crops.

Given the above, this study aimed to verify the reuse possibility of *C. ensiformis* and *C. juncea* as green manure and the influence of the vegetation cover after phytoremediation of soil contaminated with sulfentrazone regarding the growth and development of *Pennisetum glaucum*.

Material and Methods

Two greenhouse experiments were carried out. One of them was conducted with Sunn hemp (*C. juncea*) and the other with jack bean (*C. ensiformis*), two green phytoremediation fertilizers species of the sulfentrazone herbicide in the soil (Madalão et al., 2012a; b; 2013; 2017). Both experiments were conducted in an identical form.

The experiments were assembled in a 4 x 3 factorial, in a randomized block design, with four replicates. The first factor was composed from the combination of four population densities of the phytoremediation species and the second was from three sulfentrazone doses (0, 200 and 400 g ha⁻¹), totaling 12 treatments.

For *C. juncea*, the densities were 0, 60, 120 and 240 plants m^{-2} and for the *C. ensiformis* densities were 0, 10, 20 and 40 plants m^{-2} . For the two species, these densities correspond to zero, 1, 2, and 4 times the recommended density in the green manure.

As substrate for growing the plants, soil samples collected in an area with no history of herbicide application in the 0-20 cm depth, with a sandy loam texture, sieved in a 4 mm mesh and later characterized physically and chemically (Table 1) were used. This characterization served as basis for the correction and fertilization of the pots, aiming at a good development of the evaluated species as phytoremediation species, and performed based on the recommendation of Prezotti et al. (2007) for the bean crop.

Both the fertilizers and the respective doses applied per pot were carried out according to the recommendations of Prezotti et al. (2007), being them: 6.54 g of dolomitic lime (mixed into the soil one week prior to filling the pots); 2.22 g of nitrogen; 11.11 g of phosphorus; 2.5 g of potassium; 0.046 g of boric acid; 0.052 g of copper sulfate; 0.077 g of ferrous sulfate; 0.112 g of manganese sulfate and 0.019 g of ammonium molybdate.

After the soil preparation, we placed it in pots with 0.046 m^2 area and 24 cm of height, coated with polyethylene film, in order to maintain them as a closed system. 10.0 kg of the substrate were used per pot, weighed individually as they were filled. The irrigation of the plants was by adjusting the soil moisture to approximately 80% of the field capacity, followed by sulfentrazone application with the aid of a CO₂ pressurized sprayer, equipped with two TT 110.02 tips spaced 0.5 m from each other, and calibrated for 150 L ha⁻¹ application of herbicide spray.

The sowing of *C. ensiformis* and *C. juncea* species was carried out eight days after sulfentrazone application. Usually,

Table 1. Chemical and textural composition of the arable layer	r (0-20- cm) of the soil us	sed in the experiment
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Granulometry g kg ¹									
	Clay Silt			Sand		Textural classification			
	120		104		776			Sandy loam	
Chemical Analysis									
рН	Р	K+	Ca ²⁺	Mg ²⁺	H+AI	Al ³⁺	CEC	V	ОМ
H ₂ O	(mg	dm-3)	(cmol _c dm ⁻³)				(%)	(dag kg ⁻¹)	
5.2	90	17	0.6	0.2	2.4	0.4	3.2	26.0	2.0

the sowing would happen at the end of the main crop cycle, in order to avoid intoxication of the crop in succession. However, in this research, we opted for a short interval between application and sowing in order to expose the phytoremediation agents to readily available doses, without adsorption effects due to the longer residence time of the molecule within the soil (Ferraço et al., 2019).

Seven days after the emergence (DAE), the thinning was performed, leaving the number of plants per pot corresponding to each treatment. Irrigations were carried out three times a day to maintain soil moisture at 60% of field capacity (FC). The value of the FC was determined in a preliminary test to the implantation of the experiment, using the proposed methodology by Casaroli & Jong Van Lier (2008), considering a water content rate of decrease $|d\theta/dt| = 0,001 d^{-1}$.

At 75 DAE, the plants were collected separating the aerial part and root; the material was then frozen at about -20 °C for further bioassay and chromatographic determinations.

The samples of *C. juncea* and *C. ensiformis* plants were used at this stage to determine the sulfentrazone accumulation in the plant (aerial part and root) using high performance liquid chromatography (HPLC).

Chromatographic analysis was performed by HPLC means, using a Waters 2695 model equipment with UV/DAD detector, auto-injector and oven for columns. The chromatographic condition was optimized and validated as described by Ohmes & Mueller (2007), and it was used: mobile phase consisting of acetonitrile - 0.1% phosphoric acid (60:40); RP-18 column, 250 x 4.6 mm, 5 μ m; flow of 1 mL⁻¹min; 10 μ L injection, detection at 220 nm and column temperature at 30 °C. The standard solution was prepared from the sulfentrazone standard with a declared content of 99.8% at a concentration of 0.01 mg mL⁻¹.

For the determination of sulfentrazone residues in the aerial part and root of phytoremediation plants, the plant material was subjected to extraction by maceration. The previously frozen samples were removed from the freezer and, after reaching room temperature, were dried on paper towels and weighed in analytical balance at the 1:10 proportion (Barbosa, 2001; Serafim et al., 2007), with 5 g of vegetable material to 50 mL of methanol. Afterwards, the mixture was macerated until complete dispersion of the sample. After the maceration step, the samples were filtered on filter paper, and subsequently centrifuged for 15 minutes at 3.200 (rpm) for separation. Part of the supernatant extract was removed with the aid of a syringe and filtrated at 0.45 μ m on a Millipore[®] PTFE membrane into 1.5 mL vials, which were then subjected to chromatographic analysis.

The qualitative analysis, with the identification of sulfentrazone in the extracts, was held by comparing the retention time obtained in the chromatograms of the standard solution and from each sample.

The quantitative analysis was held by comparing the peak areas between the standard solution and the sample solutions. The herbicide amount found in soil, aerial part and root samples was calculated in mg kg⁻¹ of sample.

Another part of the frozen vegetal material was triturated and later diluted in water, constituting a vegetal extract that was applied in 300 mL plastic pots containing washed sand as substrate. The aerial part of the phytoremediation plants was used as an extract to provide sulfentrazone for the substrate solution, and consequently for plants grown in succession (bioassay).

The amount of aerial part extract was determined according to each species (*C. juncea* and *C. ensiformis*) and based on the results obtained by Cavalcante et al. (2012). After preparing the substrate, the bioindicator plant for the sulfentrazone presence was sown (millet – *Pennisetum glaucum* var. ADR7010).

At 15 and 30 days after emergence (DAE) of millet, the phytotoxicity was visually evaluated by using a percentage scale, ranging from 0 to 100%, where 0 (zero) corresponds to absence of symptoms and 100% to the death of plants (SBCPD, 1995); and the height of plants, by using a graduated scale, with the apical meristem as reference. At 30 DAE, the millet plants were cut close to the substrate. Subsequently, the plant material (aerial part and roots) was placed in a forced air circulation oven ($70 \pm 2^{\circ}$ C) until reaching constant weight, thus determining the dry matter of the aerial part and the root.

The use of this methodology aims to detect if the herbicide accumulation in plant tissues occurs in a phytotoxic form, after absorbing it from the soil, affecting sensitive plants in the eventual maintenance of phytoremediative plants in the field, as source of stubble or then incorporated for organic matter addition.

After data collection and tabulation, these were submitted to analysis of variance. The significant effects analysis of the population density of the phytoremediate species within each dose of the herbicide was held by analysis of regression, and the coefficients of the tested equations by the t test were at 5% of significance, whose equations were fitted according to the variable in question and its respective biological behavior. The significant effects of the sulfentrazone doses in each population density were verified by Tukey test means at 5% significance due to the insufficient number of levels for the regression equations fitting.

Results and Discussion

Figure 1 shows the recorded sulfentrazone peaks at 5.1 min. of the chromatographic injection run time, with the presence of the herbicide in the standard and in the leaf samples. In the roots, the sulfentrazone was not detected and, therefore, its data are not displayed.

It was not possible to perform the statistical analysis for the results of the sulfentrazone analytical determination in the plants, therefore, the means of the values found are displayed instead (Tables 2).

Regardless of the evaluated sulfentrazone doses and the densities of *C. juncea*, no residual herbicide was found in the aerial part, indicating that it may be degraded (phytodegradation). Similar results were found in the aerial part of *C. ensiformis* at all evaluated densities at the 200 g ha⁻¹ dose. When 400 g ha⁻¹ of sulfentrazone was applied in the soil, the herbicide was quantified in the aerial part of *C. ensiformis*



Figure 1. Chromatograms of the tested samples contaminated by sulfentrazone.

Table 2. Sulfentrazone quantity found in the aerial part of the phytoremediative plants *Crotalaria juncea* and *Canavalia ensiformis* grown at four different population densities, in a soil contaminated with three levels of the herbicide.

Deces	C. ensiformis plants m ⁻²								
(a ha-1)	10		2	40					
(g lia -)	μg of sulfentrazone for g of leaves								
0	-	-	-	-	-	-			
200	-	-	-	-	-	-			
400	0.0905	(48.91)*	0.0783	(42.32)*	**	**			
	<i>C. juncea</i> plants m ⁻²								
	6	60	1	240					
0	-	-	-	-	-	-			
200	-	-	-	-	-	-			
400	-	-	-	-	-	-			

^{*}Quantified percentage value regarding that applied in the soil.

at the densities of 10 and 20 plants m^{-2} . At the density of 40 plants m^{-2} , the herbicide was detected; however, it was not possible to quantify it (Table 2) as it was found in very low concentration in the plants.

The values found in the aerial part of *C. ensiformis*, at the densities of 10 and 20 plants m⁻², corresponded to almost 50% of the total applied in the soil, which is equivalent to a very significant phytoextraction (Table 2). When the herbicidal molecule is detected in the aerial part of phytoremediate species, it can be inferred that it was absorbed from the soil and translocated to the aerial part (phytoextraction) and then subsequently accumulated (phytoacumulation). Based on the results, it can not be determined whether part of the extracted herbicide was degraded, or whether the plant absorbed only the detected amount. However, we can affirm that the molecule was not completely mineralized or even degraded (metabolized), otherwise the sulfentrazone molecule would not be detected in the aerial part.

With the density increase, the sulfentrazone residue in the aerial part decreased to the point where in the highest density of *C. ensiformis* the herbicide was detected, but not quantified. This result can be attributed to the fact that, with a greater number of plants and their corresponding higher biomass produced; there was a dilution effect of the herbicide on the aerial part.

P. glaucum cultivated in substrate that received leaf extract from *C. ensiformis* and *C. juncea*, previously cultivated in soils with the sulfentrazone presence, showed no symptoms of intoxication to this herbicide (Figure 2).

Regarding the variables height, aerial part and root dry matter of *P. glaucum*, at 30 DAE, when comparing the sulfentrazone doses, no differences were observed between the treatments with the presence of the extract from *C. ensiformis* or *C. juncea* and in their absence (Tables 3 and 4). Similar results were obtained by Procópio et al. (2007) when field evaluating the phytoremediation of the herbicide trifloxisulfuron-sodium by *Mucuna pruriens* and *C. ensiformis*, and concluded that the stubble maintenance from the green manure species during the bean cycle (indicative species) did not affect the plants development and did not cause losses in



Figure 2. Comparison between *Pennisetum glaucum* plants at 30 days after emergence, after receiving leaf extract from the phytoremediative species *Canavalia ensiformis* (A) and *Crotalaria juncea* (B), previously cultivated in soils with sulfentrazone presence in the 400 g ha⁻¹ dose, at sowing densities of 0, 10, 20 and 40 plants m⁻² and 0, 60, 120 and 240 plants m⁻², respectively.

^{**}It was possible to detect the sulfentrazone presence, but not to quantify it.

Table 3. Height, aerial part and root dry matter of *Pennisetum glaucum*, cultivated in sand containing aerial part extract from *Canavalia ensiformis*, obtained after phytoremediation in four population densities, in a soil contaminated with three levels of the sulfentrazone herbicide.

Sulfentrazone	Number of <i>C. ensiformis</i> plants m ⁻²				
doses (g ha-1)	0	10	20	40	
	Height (cm) at 15 DAE*				
0	2.64	3.58 a	3.55 a	3.75 a	
200	-	3.32 a	3.76 a	3.62 a	
400	-	3.55 a	4.25 a	4.21 a	
C.V. (%)		14	.63		
		Height (cm) at 30 DAE		
0	3.96	5.28 a	5.31 a	5.12 a	
200	-	5.42 a	5.98 a	5.83 a	
400	-	5.68 a	6.02 a	6.03 a	
C.V. (%)	12,30				
	Aeri	al part dry m	iass (g) at 30	DAE	
0	0.03	0.09 a	0.10 a	0.12 a	
200	-	0.09 a	0.12 a	0.13 a	
400	-	0.09 a	0.10 a	0.13 a	
C.V. (%)	36.86				
	Root dry mass (g) at 30 DAE				
0	0.04	0.10 a	0.11 a	0.11 a	
200	-	0.09 a	0.10 a	0.10 a	
400	-	0.09 a	0.10 a	0.13 a	
C.V. (%)	61.85				

^{*}Means followed by the same letter in the column do not differ among themselves by the Tukey test at 5%. *DAE = days after emergence.

crop yield, indicating that the stubble permanence on the soil surface does not promote recontamination of the area. On the other hand, Nascimento et al. (2015) detected the presence of the herbicide picloram in the leaf extract from *Panicum maximum* cv. Tanzania and *Eleusine coracana* (finger millet) at phytotoxic levels for soybean plants, which forces the biomass removal of these grasses from the area in question, limiting the phytoremediation for this herbicide.

The results obtained in this work indicate that the phytoremediate species (*C. ensiformis* and *C. juncea*) can be reused as green manure after phytoremediating the sulfentrazone in the soil, making the insertion of the same in a program of rotation or succession of cultures feasible. This information is of utmost importance when thinking of the agronomic use that phytoremediation of herbicides may have. Madalão et al. (2017) evaluated the phytoremediation potential of *C. ensiformis* in the field and concluded that this species is efficient in remedying sulfentrazone. They also emphasized that it is not necessary to remove the residuals of the remediation species before planting the bioindicator species.

Among the strategies that the plants use to tolerate PROTOX inhibitor herbicides, the minimal absorption and translocation of the herbicide, as well as its sequestration, increased concentration of the mitochondrial PROTOX enzyme, which serves as a reducer for the protoporphyrinogen excess in the cytoplasm (Higgins et al., 1998, Matsumoto et al., 1999), and the rapid metabolism (Vaughn & Duke, 1991). Velini et al. (2005) checked that configurations of the PROTOX

Table 4. Height, aerial part and root dry matter of *Pennisetum glaucum*, cultivated in sand containing aerial part extract from *Crotalaria juncea*, obtained after phytoremediation in four population densities, in a soil contaminated with three levels of the sulfentrazone herbicide.

Sulfentrazone	<i>C. juncea</i> plants m ⁻²				
doses (g ha-1)	0	60	120	240	
	Height (cm) at 15 DAE*				
0	2.99	3.97 a	4.43 a	4.55 a	
200	-	3.16 a	3.25 a	3.76 a	
400	-	3.68 a	4.33 a	4.65 a	
C.V. (%)		15.	.97		
		Height (cm)	at 30 DAE*		
0	4.22	6.85 a	6.90 a	7.31 a	
200	-	6.27 a	6.52 a	6.70 a	
400	-	6.27 a	6.52 a	7.20 a	
C.V. (%)		12.	.74		
	Aeri	al part dry m	ass (g) at 30	DAE	
0	0.03	0.19 a	0.25 a	0.26 a	
200	-	0.14 a	0.18 a	0.27 a	
400	-	0.20 a	0.25 a	0.25 a	
C.V. (%)	32.44				
	Roots dry mass (g) at 30 DAE				
0	0.05	0.19 a	0.30 a	0.34 a	
200	-	0.11 a	0.18 a	0.29 a	
400	-	0.14 a	0.20 a	0.24 a	
C.V. (%)	59.88				

*Means followed by the same letter in the column do not differ among themselves by the Tukey test at 5%. *DAE = days after emergence.

enzyme or promoters that would allow different levels of expression might allow genotypes that are more tolerant to the herbicides that act in inhibiting this enzyme.

When evaluating the height and dry matter data from aerial part and root of *P. glaucum* at 15 and 30 DAE, regarding the planting density of *C. ensiformis*, we observed that the height increased according to the densities increase of the remedy. The same happened for the aerial part and root dry matter evaluated at 30 DAE (Figure 3), with their regression equations displayed in Table 5. This fact may be due to the increase in the population density of the phytoremediate species that resulted in the greater absorption of the nutrients present in the soil, which may have returned via extract to the used substrate (washed sand), and thus favoring a greater growth of the millet. Similar results were also observed by Ferraço et al. (2017) when evaluating the effect of the population density of *C. ensiformis* on the phytoremediation of soil contaminated with sulfentrazone.

Similar to what was observed with *C. ensiformis*, at 15 and 30 DAE, *P. glaucum* grown on substrate containing extract from the *C. juncea* aerial part presented a height increase according to the remediation species densities increase. The same situation was observed for the aerial part and root dry matter at the 30 DAE (Figure 4), with their regression equations displayed in Table 6. Oliveira et al. (2014) studied the phytoremediation of soil contaminated with sulfentrazone by the species *C. ensiformis* and *C. juncea* using *P. glaucum* as bioindicator and concluded that both species are efficient in remedying soils contaminated with sulfentrazone.



Figure 3. *Pennisetum glaucum* height at 15 and 30 days after emergence (DAE), aerial part and root dry matter at 30 DAE, cultivated in washed sand containing extract from *Canavalia ensiformis* aerial part, obtained after phytoremediation in four population densities, in a soil contaminated with three levels of the sulfentrazone herbicide.



Figure 4. *Pennisetum glaucum* height at 15 and 30 days after emergence (DAE), aerial part and root dry matter at 30 DAE, cultivated in washed sand containing extract from *Crotalaria juncea* aerial part, obtained after phytoremediation in four population densities, in soil contaminated with three levels of the sulfentrazone herbicide.

In cases such as that of the picloram herbicide, phytoextraction and consequent phytoaculation implies in phytotoxicity to sensitive species cultivated on the stubble containing this herbicide, which invalidates its reuse and makes the application of phytoremediation harder (Nascimento et al., 2015). However, regarding the sulfentrazone, with the obtained results by the bioassay, we can infer that it is possible to keep the green manure stubble in the phytoremediated area (Figures 2, 3 and 4 and Tables 2, 3 and 4), agreeing with the results found by Procópio et al. (2007) about the reuse of the biomass from the *Stizolobium aterrimum* and *C. ensiformis* species, regarding the herbicide trifloxysulfuron-sodium.

Conclusions

As for the phytoremediation mechanisms of sulfentrazone, we can affirm that this herbicide was absorbed by the plants of *C. ensiformis* and not only just immobilized in the soil.

Rhizospheric action as a probable detoxification mechanism of sulfentrazone has not been proven by this assay; however, it is possible that *C. juncea* has used this strategy in order to remediate the herbicide in the soil, emphasizing phytostimulation and rhizotransformation as the most probable biological mechanisms.

The sulfentrazone molecule was absorbed from the soil and translocated to the aerial part (phytoextraction), the accumulated one (phytoacumulation), but was not completely mineralized by *C. ensiformis*, a behavior that would be desirable for phytoremediation of agricultural areas, although it did not result in posterior toxicity.

Further tests are necessary in order to investigate the sulfentrazone destination and its possible metabolites and the involved mechanisms in the phytoremediation of this herbicide.

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